

The tester plants in part II of table 19 were also used to determine if the uniformly pigmented plants derived from uniformly pigmented kernels

appearing on ears / produced by crosses of plants carrying the 5720 state of a_1^{m-1} (see table

18 for selections) carried a stable mutant of state ~~5720~~ that would not

respond to Spm, in any manner. These plants had ~~a derivative of~~ state

derivative 5720 ~~a_1^{m-1}~~ and Sh_2 in one chromosome 3 and $a_1^{m-1} sh_2$ in the homologue.

If Spm were also present in them, to which the mutant ~~was~~ ^{did} not responding,

then its presence would be ~~clearly~~ revealed by the appearance of variegated

a_1^{m-1} (state 5719A-1), kernels within the $/sh_2$ carrying ~~xxxxxxxx~~ class on ears produced by

^{of them} crosses with the tester plants in part II of table 19. This type of test,

as well as some others that will be described later, indicated the stability

of the mutants derived from state 5720, for some of the plants ^{such a mutant} ~~carrying them~~ ^{with}

^{caused} also had Spm.

The tester plants in Part III of table 19 were particularly useful

for introducing the Spm element(s) in them into kernels on ears produced

by plants that were derived from pale, Sh_2 ~~kernels~~ ^{state} in table 18. This was

especially important when the state of a_1^{m-1} in them was not 5719A-1.

It was evident from the phenotypes of kernels on the resulting ears that
In all crosses of this type, the state of a_1^{m-1} carried in the plant being

tested would respond to the Spm delivered to the endosperm by the tester plant. If the plant being tested was a_1^{m-1} (any state but

5719A-1) $Sh_2/a_1 sh_2$ in constitution, the difference in ~~variegation~~ ^{variegation exhibited by} pattern ⁱⁿ the Sh_2 class of kernels when compared with that in the sh_2

class was always evident and this was strikingly so when the plant being tested had either state 5700A or state 5996-4. ^(non-pale for the pattern class produced) This was another test

that confirmed the unique response of each state of a_1^{m-1} to any one Spm element. It was also important for demonstrating the relation of

response of the a_1^{m-1} state in the plant being tested to the particular Spm constitution in the tester plant. ^{on one plant,} ~~If the~~ Spm were linked with a known

gene marker (Y Spm / y + in plant 6666E, table 19), ^{among} the kernels on ears

produced when its pollen ^a was used to make the cross ~~showed~~ linkage of

the variegated class of kernels ^{with Y and the pale colored class with y was expressed} ~~with this marker~~ on all ears where the

constitution ~~with respect to this marker~~ ^{was 4/4} of the plant being tested ~~would~~

~~all this to be expressed.~~ ~~Also,~~ The ratio of variegated to pale kernels

^{among} ~~in both~~ the Sh_2 ^{test among} kernels and the a_1^{m-1} ^{class} carrying sh_2 kernels reflected the

number of Spm elements that were present in the tester plant (1 Spm

in plants 6666E, 6671E, 6673F and 6690 and at least 3 in plant 6680D). ^{table 19}

Plants derived from the uniformly pale colored kernels in table 18

from some nuclei during development as a consequence of transposition, or from ^{effectiveness} ~~loss of its activity~~ as a consequence of its inactivation. If only ^{the occurrence of} ~~one~~ ^{two} one Spm were present, either of these events ~~occurring to the Spm element~~ in a nucleus would result in the appearance of pale pigment in its progeny cells. If two or more Spm elements were present in a nucleus, then an event must occur to each of them at one time, or a succession of such events must occur, first to one Spm element and then to the other, in order that a nucleus arise in which there is no Spm activity. Thus, the patterns of variegation exhibited both by kernel (see page 00) and by ~~parent~~ ^{originally present} plant may be strikingly different depending upon the Spm number in their nuclei. However, this relationship ~~is~~ ^{is not} an absolute one, ~~for~~ ^{it} also depends both upon the time during development when these events occur, and the frequency of their occurrence at any one time, and this is a property of the isolate of Spm present in a kernel or a plant. Spm elements undergo change in state that alter these properties and discussion of this will be given in sections devoted to transposition of Spm and to its inactivation. For this reason, not all kernels and plants having one Spm element in them ~~may~~ ^{will} exhibit ~~a high frequency of appearance of~~ ^{unstable} pale areas. The Spm element in them may not be ~~subject to~~ ^{undergoing} frequently occurring modification of the types described.

the mode of operation of

Discussion of ~~the~~ tests of Spm conducted during the summer of 1954, will commence with progeny of plants in culture 6629A (table 5) whose constitutions were a_1^{m-1} (state 5719A-1) $Sh_2 / a_1 sh_2$ and ^{also} considered to be Y Spm / y [^]. The silks of the ears of plants in 6629A had received ^{culture} pollen from plants of the standard tester stocks, homozygous for a_1 , sh_2 and y. ^{were selected from} Selected kernels ^{modulated y} on two of the ears of plants 6629A-1 and ~~plant~~ ^{the types of kernels selected and} 6629A-7 and ^{from} one ear of plants 6629A-4, 6629A-6, and 6629A-9. ^{are chosen in} ~~were~~ sown ^{of the} in Table 18¹. The phenotype of some kernels on the ear of the tiller of plant 6629A-7 were aberrant. Plants derived from selected kernels on this ear were grown under culture number 6675, and ~~tests~~ of them will be considered separately ~~following discussion of~~ Those conducted with progeny derived from the five other ears ^{were} grown under culture numbers 6665, 6666, 6670, 6673 and 6674.

In table 20 is given the number of plants in each of the above five cultures that were derived from kernels with yellow (Y) or white (y) endosperms among the pale Sh_2 , the variegated Sh_2 , and the colorless, sh_2 classes of kernels on the parent ears. In addition, one plant was grown from a variegated, sh_2 , Y kernel in culture 6666. Four other plants were grown from kernels whose phenotypes differed from others on the

Tests to determine

parent ears, as described in the footnote of table 20. ¹ The nature of the modification responsible for them will be considered later ~~when tests~~ ^{along with those} conducted with ~~other~~ other plants derived from similar types of kernels. ~~xxxxxxxxxxxxxxxx~~

table 20,

All 61 plants derived from the pale, Sh₂ class of kernels were ^{was the same in all but it was lighter than that appearing when A₁ is present.} uniformly pigmented, ~~and the type of pigment was that produced when Spm~~ ^{is absent.} Among the 66 plants derived from the variegated kernels, all were variegated except one. This exceptional plant was uniformly pigmented and the type and distribution of pigment was the same as that in plants derived from the pale class of kernels. Tests conducted with this plant gave no evidence of the presence of Spm in it. It was learned subsequently that loss of Spm from a nucleus, either during development of the female gametophyte or during very early embryo development, will give rise to a kernel whose endosperm and embryo differ in constitution, and

cases of this will be considered in the section devoted to transposition.

Also, inactivation of Spm early in development of a plant gives rise to the pale pigmentation of the endosperm and embryo, as indicated in the table.

Tests conducted with ~~xxx~~ plants entered in table 20 were both numerous and varied in type. Crosses were made with plants having the following constitutions: the standard tester stocks, homozygous for a₁ and the sh₂, / three different ^{categories} ~~types~~ of tester plants whose constitutions are given in table 19, and with other plants having states of a₁^{m-1} not

modification of Spm

most types of test-cross

Table 20

entered in table 19. The ~~tests~~ conducted with each plant will not be

^{here} listed but in order to illustrate the scope of ^{these} such tests, those conducted

with each ~~of the~~ plants in cultures 6665 and 6666 are given in tables 21 and

22. ^{for plants derived from the S₁ crossing} As indicated previously, ~~these plants were derived from selected~~

^{for the S₁ crossing} kernels on the main ear and on the tiller ear of plant 6629A-1 (see table

5 for kernel types on each of these ears). Including the many ears obtained

from crosses conducted with plant 6666E, whose constitution is given in

table 19, over 130 ears were produced from test crosses conducted with the

^{6665 and 6666} plants in these two cultures. Because transposition of S_{pm} may occur in

some cells during development of a plant, and result in loss of S_{pm} or

in change in its number in some somatic nuclei, the kernel types on each

^{as well as} ear ~~and~~ the ratio of types had to be considered with this possibility in

mind. Nevertheless, on many ears produced by one type of test-cross,

both the phenotypes of kernels and the ratios of these were often much

^{in the tables} alike. Therefore, ^{and} data obtained from such ears ~~may~~ be summed. On

some ears, however, the ratio of phenotypes did not agree with that given

by the majority of ears, and the cases where this was noted will be

^{these} indicated, ^{from a practical} as deviation often proved to be of considerable significance, ^{that are} furthering an understanding of S_{pm} behavior.

There were two objectives for making the test-crosses entered in

parts I and II of table 21. One was to establish whether or not Spm were present in a plant, and the second was to determine if the a_1^{m-1} in the plants not having Spm ^{were} ~~would be~~ capable of responding to it.

If Spm were absent, no variegated kernels should appear on any of the eight ears produced by the test-cross with the standard stock that was homozygous for a_1 and sh_2 , and as shown in ^{the first line of} ~~column 2 of~~ table 23, none appeared. Also, if these plants are crossed by plants of tester type I or II, table 19, no variegated kernels should appear on any of the resulting ears, and as lines 2 and 4 of table 23 indicate, none appeared.

However, if tester plants of type III, table 19, are used in crosses with these plants, then some variegated kernels in the Sh_2 classes should appear if the a_1^{m-1} in these plants is capable of responding to Spm. These kernels should have pigmented areas in a colorless background. The

Spm delivered by the male parent plant to the endosperms of kernels on

the ear of the tested plant could allow the a_1^{m-1} in these ^{plants} ~~latter~~ plants

^{the female parent} to exhibit its mode of ^{response} ~~reaction~~ to ~~the presence of~~ Spm. The ratio of

pale to variegated kernels in the Sh_2 class would depend upon the Spm

constitution of the tester plant. In all of the many crosses to pale-^{plants}

^(part II, table 19) plants conducted with plant 6690, and with ^{repeated among them} all states of a_1^{m-1} , a ratio

^{of approximately 1 pale to 1 variegated kernel} appeared on each ear, ^{and} ~~as~~

line 5 of table 23, ~~shows~~, this ratio appeared in the Sh_2 class of kernels ^{cultured in}

(part 1, table 1a)
Tester plant 6680D¹ also was used in crosses to many plants with the pale

phenotype ^{to many which} and ~~those with~~ different states of a_1^{m-1} ^{was repeated.} ~~included among them.~~

On all ears so produced, nearly all the Sh_2 carrying kernels were variegated -- pigmented areas in a colorless background. The data obtained from the cross of this plant to plant 6665B-5, line 6, table 23, is an illustration of this. The pollen of plant 6666E, ~~which was used~~ was used on silks of ears of three of the pale, y/y plants in part II of table 21. Its pollen likewise was placed on the silks of ears of many other plants with a pale phenotype and on all ears a ratio approximating 1 pale to 1 variegated kernels appeared. When the plants being tested were y/y in constitution, linkage of the variegated class with Y and the pale class with y was always expressed among the kernels on an ear. It ^{was} ~~could be~~ concluded that plant 6666E carried on ^{that the} ~~it~~ was located in its Y bearing chromosome 6. Among the Sh_2 kernels on each of the three ears contributing to the data entered in line 7 of table 23, this likewise ^{linkage} was exhibited. ^{Among} ~~In~~ the 413 kernels in the pale Sh_2 class, 160 were Y and 253 were y. Among the 306 Sh_2 kernels in the variegated class, 186 were Y and 120 were y.

It is now necessary to show that the a_1^{m-1} in each of the tester plants of type I, table 19, is capable of responding to the presence of Spm. The capacity of the a_1^{m-1} in each of these plants to respond to Spm was strikingly illustrated by the phenotypes of kernels appearing on ears of plants derived from the colorless, sh_2 classes of kernels, table 20, when pollen from a tester plant of Type I was placed on the silks of ~~an~~ ear^s of these plants. ~~The crosses made with each such plant in cultures 6665 and 6666 are entered in table 22.~~ Although it could be assumed in advance that no evidence of Spm would be ~~detected in~~ ^{obtained from} test-crosses conducted with the pale pigmented plants in parts I and II of table 21, and that evidence of its presence would be obtained from tests conducted with the variegated plants, entered in parts III and IV of ^{this} ~~table 21~~, no such assumptions could be made with regard to any one plant derived from the colorless, sh_2 kernels. They do not have a_1^{m-1} and no anthocyanin develops in either kernel or plant. It would be expected, however, that Spm would be present in some of them but absent in others. Since, in ^{the parent} ~~of these in the culture entered in table 20,~~ plants ~~6629A-1~~, Spm was considered to be located in the Y-bearing chromosome 6, it could be anticipated that Spm would be present in more plants ^{colorless, sh_2} ^{on this ear} ^{colorless, sh_2} derived from the Y class of kernels than in those derived from the y

class, and this proved to be true.

Tests of plants derived from the colorless, sh_2 classes of kernels were considered to be of first importance for verification of the mode of action of Spm and the response of each state of a_1^{m-1} to ~~it~~^{any one Spm.}. Therefore, many tests were conducted with these plants, not only with those in cultures 6665 and 6666, entered in table 22, but also with all plants derived from the colorless class of kernels that are entered in table 18. The evidence obtained from tests conducted with them bears directly on that already discussed, and also on that obtained from tests of plants entered in parts III and IV of table 21, not yet discussed. For this reason, the tests conducted with plants derived from the colorless, sh_2 kernels in cultures 6665 and 6666 will be considered now.

An attempt was made to test each of the 31 plants derived from the colorless, sh_2 Y class of kernels and from the 25 plants derived from the colorless, sh_2 y class of kernels in cultures 6665 and 6666 (table 20). This was successfully accomplished with 30 of the former and with 24 of the latter. The types of test conducted with each plant are entered in table 22. The plants in this table are placed under four sub-headings: ~~that~~ that were Y/y and in which Spm was found to be present (part I) or

absent (Part II) and those that were y/y and having Spm (part III) or in which Spm was found to be absent (part IV). The first ear of the main stalk of these plants received pollen from one of the ~~Type~~ I tester plants entered in part I of table 19. The majority of the Y/y plants ~~xxxxxxxx~~ *of this table* were crossed by plants in culture 6638A. All plants in culture 6638A were homozygous for state 5718 a_1^{m-1} , for Sh_2 and also for the recessives, y, pr, and wx. None of them had Spm, as the many tests conducted with each soon illustrated. Similar tests, equally extensive, were conducted with the plants in culture 6641, whose constitutions are entered in lines 2 and 3 of table 19. Each plant was homozygous for state 5719A-1 a_1^{m-1} and none of them had Spm. / Pollen collected from plants in this culture was placed on silks of the first ear of the main stalk of some of the plants entered in table 22. Also, as this table shows, pollen collected from pale-pigmented plants having other states of a_1^{m-1} (states 5719A-2, 5700A, 5714F, and 5996-4) was ~~also~~ used on silks of other ears of some of these plants.

As table 22 indicates, with some plants, more than one ear per plant was used in making a test cross, and the pollen parents differed with respect to the state of a_1^{m-1} present in them. The purpose of this multiple test of a single plant is evident. If no Spm were present in the plant, then no variegated kernels should appear on any ear it produces.

If, however, Spm were present, then variegated kernels could be expected to appear on each ear and the pattern of pigmented spots in a colorless background among all of them on any one ears should be similar, -- that controlled by the particular state of a_1^{m-1} that was introduced by the pollen parent. This expectation is based on the interpretation previously outlined of the individual response of each state of a_1^{m-1} to any one Spm. Another relationship could also be anticipated to appear among the kernels on these ears. It could be expected that in the plants derived from the colorless, sh_2 , Y, kernels, Spm would be present in some, and in most of them it would be carried in the + bearing chromosome 6. Thus, on ears of these plants, linkage of the variegated class of kernels with Y and the pale class with y could be anticipated, regardless of which state of a_1^{m-1} the pollen parent had introduced. The expectations outlined above were fulfilled in tests of plants in cultures 6665 and 6666, and also, it may be added, in many tests of similar type that were conducted with plants in other cultures.

As stated earlier, the plants in table 22 are sorted into four groups according to their constitutions with respect to the alleles of Y and to the presence or absence of Spm in them, based on results obtained from the above described tests of each. On none of the ears produced

by plants entered in parts II and IV of this table did variegated kernels appear. Only the pale pigmented class of kernels appeared, and on each ear, the pigment in these kernels was ~~of~~ the type that is characteristically produced in the absence of Spm by the state of a_1^{m-1} introduced by the pollen parent. In contrast, on all ears produced by the plants entered in parts I and III of table 22, variegated as well as pale-colored kernels appeared. On any one ear, the type of expression of variegation among all kernels exhibiting it was the same. Similarly, the pattern of variegation appearing among the variegated kernels on ^{plants} different ears was the same ^{used in the cross} provided that the pollen parent in ~~each case~~ [^] had introduced the same state of a_1^{m-1} .

The ratio of variegated to ~~pale~~ colored kernels on these ears also was instructive, for ~~it~~ indicated the Spm number in each tested plant. The

ratio of these two classes of kernels on ears produced by the test crosses conducted with the 15 plants entered in part I of table 22 are given in table 24. It may be seen that on the ears produced by 12 of these 15

plants, one Spm was present and it was carried in the Y bearing chromosome

(Part I, table 24) in each plant. On the ears produced by plant 6666G-4, part II, table 24,

the data were too few to indicate with certainty linkage of Spm with Y

although ^{they are} ~~it is~~ suggestive of this. However, the test-cross conducted

with plants 6665G-16 and 6665G-21, Part III of table 24, did not give ^{obtain} ~~good~~ evidence of linkage of Spm with Y. In order to ^{ascertain} ~~be certain~~ of the location of Spm in these two plants, tests were conducted the following year with plants derived from the variegated, Y class of kernels on each of these ears. Tests of the progeny of plant 6665G-21 showed that the Spm in it was not linked with Y. Similar tests conducted with the progeny of plant 6665G-16 suggested that the Spm in this plant was still carried in chromosome 6 but at a location farther removed from Y than in the parent plant, 6629A-1, ^{or} ~~and~~ in sister plants entered in part I of table 24.

Among the 24 tested plants derived from the colorless, sh_2, y kernels parts III ~~and~~ ^{IV}, table 22, only 6 had Spm. One test cross ear per plant was obtained from 3 of these 6 plants and 2 test cross ears were ^{from plant} ~~were~~ obtained from the remaining 3. The types of crosses conducted with these plants are shown in part III of table 22 and the ratio of pale to variegated ^{among those that carried a_1^{m-1}} kernels/on each ear is given in table 25. The ratio of pale colored to variegated kernels on each of the 9 ears obtained from tests of these six plants indicated the presence of one Spm in all six of them to which states 5718, 5719A-1, 5719A-2 and 5996-4 of a_1^{m-1} responded, each in its own manner.

of plants

The tests/derived from the colorless, sh_2 classes of kernels

entered in table 22 clearly expressed the effectiveness of the test method for revealing Spm constitutions in these plants and for demonstrating the individuality of response of each state of a_1^{m-1} to the Spm that was present in any one plant. With this in mind, consideration may now be given to tests conducted with the sister plants that were derived from the variegated, Sh_2 kernels. The ^{test}/crosses that were conducted with each of them are entered in parts III and IV of table 21. Those conducted with ~~the~~ plants derived from the variegated, Sh_2 , Y class of kernels, part III of table 21, /will be considered first.

Of the 17 tested plants in part III of table 21, 15 had one Spm *from the ratio of kernel types on the ear each produced, it was evident that of these plants* and it was carried in the Y bearing chromosome 6 in each. *(The plants* ^{in cultures 6665 and 6666 indicated} ~~having this constitution are entered in column 3 of table 21.~~) The

kernel types on the test-cross ears produced by these plants are given in

table 26. ^{ratio of kernel types on the 6666C-1} The/one ear obtained from plant did not give evidence of

linkage of Spm with Y. ^{the following year} However, tests conducted [^]with plants derived

from the variegated, Sh_2 , Y kernels on this ear indicated that the Spm

in it was carried in the Y bearing chromosome 6. ~~in this plant.~~ Similarly,

the ratio of kernel types on the first ear of plant 6666C-2, part II,

table 26, did not give evidence of linkage of Spm with Y ^{kernel types on} whereas the tiller [^]ear of this plant, part I, table 26, clearly indicated such linkage.

Therefore, tests were conducted ~~the~~ following year with plants derived from the variegated, Y kernels on the ear of the main stalk of this plant and these established the presence of Spm in the Y bearing chromosome ~~thus part of~~ ^{and also in the other.}

6 in plant 6662C-2. The ratio of kernel types on the two test-cross ears obtained from plant 6665E-10, part I, table 26, differed from that appearing on all other ears. There were 103 pale-colored, Sh_2 kernels of which 45 were Y and 58 were y, and 234 variegated Sh_2 kernels of which 119 were Y and 115 were y. Obviously, the Spm in this plant was not linked with Y. Two Spm elements could be present in it or one Spm element, could be present and carried in the chromosome 3 with a_1^{m-1} . In order to determine the Spm constitution in this plant, tests were conducted the following year with plants derived from the variegated kernels on each of the two ears of plant 6665E-10 and these ^{tests} established the Spm constitution of this plant. It had 2 Spm element, neither of which was linked with Y nor with a_1^{m-1} .

The ratio of kernel types appearing on the two test-cross ears of plant 6666C-7 were aberrant (parts I and II, table 26). Both ears were partially sterile and the number of kernels on each were few. Tests conducted with 5 plants derived from ~~some of the~~ variegated kernels on the ear entered in part II of this table indicated that a burst of transposition⁵:

of Spm ~~must~~ have occurred late in development of this plant. One plant was Y Spm/y \dagger , another had 2 Spm elements, one of which was linked with Y, a third had 1 Spm element linked with Pr in chromosome 5, a fourth had 1 Spm element linked with Wx in chromosome 9, and the fifth had 1 Spm element, not linked with any of these markers. / Also, The factor responsible for sterility was not transmitted to any one of these offspring. It is however, originally probable, that Spm was carried in the Y bearing chromosome 6 of plant 6666G-7, and was linked with Y in two of the five ^{tested} progeny plants.

One Spm was present in each of the 6 plants in part IV of table 21. On each test cross ear, a ratio of one pale-colored kernel to one variegated kernel appeared among those having a_1^{m-1} in them, as table 27 indicates.

The test crosses conducted with plant 6666E, part V, table 21, were numerous, as mentioned earlier. It was quite evident from them that the Spm in it was carried in the Y bearing chromosome 6 and evidence of this was given on page 00. Pollen was collected from the main stalk of this plant and from each of its three tillers. Tests conducted with all four parts of this plant indicated that the Spm in each ^{part} was carried in the ₁ Y bearing chromosome 6.

Altogether, 93 of the 106 plants in the progeny of plant 6629A-1 that were grown in the summer of 1954 under culture numbers 6665 and 6666

were tested for ~~their~~ Spm constitution. Table 28 summarizes the Spm constitutions and locations in the plants as determined by the test of each, given in detail in the preceding pages. Similar types of tests were conducted with plants in cultures 6670, 6673, and 6674, table 20, derived from selected kernels on ears produced by other Y Spm/y + plants in culture 6629A. The results obtained from them were so similar to those already described that a detailed description of the type of test conducted with each plant need not be given here. No Spm was found to be present in the pale pigmented plants derived from the pale colored kernels. However, in all cases where Spm was delivered by means of ^{to an endosperm of a kernel on the ear} a cross^y conducted with a plant of tester type III, table 19, the a_1^{m-1} in ~~these plants~~ ^{each} was found to be capable of responding ^{ed} to the Spm delivered by the tester plant. ~~An Spm element~~ was found to be present in all variegated plants derived from variegated kernels. In most of them, one Spm was present and in the majority of these derived from the Y class of kernels, it was linked with Y. In several plants, however, the Spm element in them did not give evidence of linkage with ^{this marker} Y.

Tests of the plants derived from the colorless, sh_2 classes of kernels were as extensive for ~~cultures~~ ^{cultures} 6670, 6673 and 6674 as for 6665^A and 6666, just described. Here, also, it was found that when Spm was present in

in any one of them, it was capable of acting upon any state of a_1^{m-1} and each state, in turn, responded to it in its own individual manner. The tests conducted with these plants may be summarized briefly. Of the 59 plants derived from the colorless, sh_2 , \bar{Y} class of kernels in table 20, 56 were successfully tested. In 32 of them, ^{one}/Spm was present ~~and~~ it was carried in the Y chromosome in 30 of ~~these~~ plants. The Spm in 2 plants ~~did~~ not show evidence of linkage with Y. Of the 63 plants derived from the colorless, sh_2 , y kernels, 60 were successfully tested for Spm. It was present in only 17 of these plants and one Spm was present in each. The percent recombination between Y and Spm in the parent plants was 35.3. This same percent of recombination appeared ~~on~~ the test cross ears of ~~the majority~~ ^{most} of the progeny plants that were Y Spm/ y + in constitution, as table 29 illustrates. Ears on which the ratio was ~~exceptional~~ ^{4-5 types}, ~~as~~ described earlier, were excluded from the table.

The tests outlined in this section serve to illustrate the manner of examining Spm constitutions in plants and the effect that any one Spm will exert on any one state of a_1^{m-1} . Many of the tests included crosses between plants carrying different states of a_1^{m-1} as well as crosses to plants that were homozygous for a_1 , either with or without Spm in them. Figure 1 is constructed to illustrate the ~~xxxxxx~~ combinations of states

that were made and photo. 00 illustrates the types of kernels appearing on an ear following such ⁰⁰ ~~a~~ cross. Plants were grown from some of the

variegated kernels in which two different states were present, one in

each chromosome 3. ^{Some of the} /Tests of them were made by crosses with plants

homozygous for a_1 and having no Spm in order to investigate the segregation

of the two states to the progeny. Each state segregated from the other

~~Phases~~ in the expected manner as if each were a distinct allele of the

other. Photo. 000 of an ear produced by a cross of a plant carrying

state 5700A and Sh_2 in one chromosome 3 and state 5719A-2 and Sh_2 in the

homologue chromosome by one that was homozygous for a_1 and sh_2 will

illustrate this. Again, if, in these plants Spm were linked with a

genetic marker, this linkage was expressed in like manner by each state

among the kernels on the test cross ear, and a large number of tests of

this type were made in examining new locations to which Spm had been

transposed.

Part III

TRANSPPOSITION OF SPM IN a_1^{m-1} CULTURES

Early in the study of a_1^{m-1} it was realized that the independently located element in the system did not remain constant either in number or in location in the chromosome complement. The first clear evidence of this was given by the ratio of kernel types on ears of plants grown in the summer of 1952 (table 3). At that time, transposition of the controlling elements Ds and Ac was known and it was considered possible that the independently located controlling element in the a_1^{m-1} cultures, comparable to Ac, was also undergoing transposition. In order to determine that change in location of this element was occurring, it was necessary to examine its location in individual plants in the progeny of those in which its number and location was known. *This number and location* ~~This~~ had been determined ~~for Spm~~ in some of the plants of culture 6629 (tables 5 and 8). Therefore, *some of the* ~~extensive~~ tests of the progeny of ~~some~~ of the plants *were undertaken for this purpose* ~~this culture~~ was conducted during the summer of 1954. Those conducted with the progeny of four of the Y Spm / y + plants in culture 6629 were discussed in the preceeding section. It was shown there that in the majority of ~~Spm carrying~~ *trans* progeny ~~plants~~, only 1 Spm was present, and that in the Y / y ~~plants~~, it was carried in ~~the~~ chromosome 6 with Y and at the same location, or certainly close to this, as in the parent plants.

progeny

In a few plants, however, Spm was not linked with the Y marker, and in one of which ^{it was} linked with Y in some plants, & neither linked with Y in other plants. Several of them, two Spm elements were present instead of one, ¹ Some of

of this the cases where such was found were ^{described} and the progeny of ^{one of these} were ^{he considered later.} mentioned.

^{many} Most of the examined plants, whose ~~constitutions were considered~~

~~in the last section~~ were derived from kernels on the testcross ears of

^{in culture 6629A,}

plants entered in table 5. In addition, an ear of ^a ~~the~~ tiller of one

of them, plant 6629A-4 that was Y Spm/y + in constitution, had been

self-pollinated. Plants derived from selected kernels on this ear were

examined for the Spm constitution in each. In ^{those that carried both} ~~the~~ Y and Spm, ^{carrying}

^{with respect to the alleles of Y and to Spm} ~~plants~~, the following constitutions ^{could} be expected to appear:

(1) Y Spm/Y Spm, (2) Y Spm/y Spm, (3) Y Spm/ Y +, (4) Y Spm/y +, and

(5) Y +/ y Spm. Among the 18 examined progeny plants having both

Y and Spm, 3 had constitution (1), 1 had constitution (2), 3 had

constitution (3), 7 had constitution (4), and 1 had constitution (5).

In the remaining 3 plants, other constitutions were found. All were

Y/y. One plant had two Spm elements, neither of which was linked with

an allele of Y. Another had two Spm elements, one of which was linked

with Y. The third plant had one Spm but it was not linked with either

Y or y.

If no changes in location or in action of Spm occur in plants having two Spm elements at allelic positions in an homologous pair of chromosomes,

then all gametes each plant produces should carry Spm. On the testcross ears of these plants, all kernels would exhibit variegation. However, changes do occur and the extent of this may be judged by the proportion of pale colored kernels that are randomly distributed over the ears they produce. In the Spm/Spm plants, mentioned above, between 10 and 15 percent of the kernels on the testcross ears they produced were uniformly pale colored instead of being variegated. These kernels represent cases in which Spm had been altered, either in location or in action, in cells of the parent plant. *insert page 124a to 124b*

Examination of the progeny of individual plants of culture 6629 confirmed the Spm constitutions that had been presumed ^{*for each, based on the ratio of*} ~~from the~~ kernel types appearing on the testcross ears each produced. However, more ^{*precise*} ~~was~~ ^{*information was obtained*} ~~learned about this~~ from tests conducted with their progeny, and this is summarized in table 30. Spm apparently occupied the same position in chromosome 6 in each of the 4 Y Spm/y + plants (A-1, A-4, A-6, and A-7) whose progeny were examined. At this location Spm ^{*does*} undergoes ~~some~~ change both in location and in type of action, ~~and cases of this were mentioned in the last section of this report.~~ ^{*but*} However, the frequency of ^{*appearance*} ~~occurrence~~ of this ^{*in the progeny*} is much less than that which ^{*appears when Spm*} ~~occurs to Spm~~ when it occupies other locations in the chromosome complement, ~~and~~ ^{*given*} evidence of this will be ~~discussed~~ shortly.

The types of testcross conducted with plants grown in the summer of 1954, whose origins are given in table 18, were described in the previous section. Some of them were made to investigate the behavior of different states of a_1^{m-1} whereas others were made to determine Spm constitutions in the progeny of plants whose Spm constitution had been determined. During the summer of 1955, many testcrosses were conducted for the purpose of investigating transposition of Spm. This required determining the presence or absence of Spm in each kernel on an ear produced by a plant whose Spm constitution ~~it~~ was ~~desired~~ to be determined. For this purpose, it was necessary to use states of a_1^{m-1} that give very ~~few~~ clearly defined patterns of mutant spots in the presence of Spm. Therefore, the tester stocks that were used in the summer of 1955 were homozygous for either state 5718 or for state 5719A-1. With each, small spots of the A_1 type pigment appear in a colorless background when Spm is present. In the absence of Spm, state 5718 gives only very faintly pigmented kernels. For tests requiring distinctions between the Pr and pr phenotypes in the pale class of kernels ^(no Spm) ~~tester stocks~~ ^{carrying state 5719A-1} usually were used. This was done because in the absence of Spm, the intensity of pigment in ^{kernels} ~~the kernels~~ is deep enough to allow distinctions to be made between the Pr and pr

phenotypes. With state 5718, such distinctions are difficult because of the faintness of pigment in them.

The state of a_1^{m-1} in the plants tested was either 5719A-1 or 5718 unless otherwise stated. The use of these states was important because the frequency of occurrence of germinal mutation is very low for each. However, occasionally a sector will appear on an ear of a plant having state 5719A-1 in which all of the kernels within it exhibit the same mutant phenotype, indicating that a mutation to or toward A_1 had occurred in a cell during development and that its progeny cells were included in a sector on the ear. Such sectors on ears appear only very rarely. In the tables given in this part of the report, kernels having germinal mutations were not included. This was done to allow the data to be presented in a simple form whereby the ratio of the pale class of kernels (no Spm) to the variegated class (Spm present) may be read rapidly. To be meaningful, inclusion of germinal mutations in these tables would also require reference to the state of a_1^{m-1} that was present in each plant, to whether or not it was homozygous for this state or heterozygous with a_1 , or whether state 5719A-1 was present in few kernels with a one chromosome 3 and state 5718 in the homologue. Since/germinal mutations ~~is low for either state,~~

mutation appeared on the testcross ears, it was considered needless to complicate the presentation, both in the text and in the tables, by detailed reference to the constitution of each plant with regard to the state of a_1^{m-1} it carried.

In the discussion that follows, the term "testcross" will be used frequently. Unless otherwise stated, it will refer to a cross made with a plant whose Spm constitution is to be determined by one that is homozygous for state 5718 or state 5719A-1 of a_1^{m-1} , for Sh_2 , and for pr , y , and wx , and in which no Spm is present.

The number of progeny tests that were made for the purpose of examining Spm constitution in ^{each plant} ~~them~~ is large. In describing each of these tests it will be necessary to refer to culture numbers under which the different progenies were grown, and to plant numbers within a culture. To lessen confusion that could arise from use of such numbers in the text, a chart was prepared, figure 2, that gives the culture number of the plant whose progeny was tested, the constitution of this plant, and the culture number of its progeny along with the table number that illustrates the Spm constitution in the plants of its progeny. Culture numbers of plant grown in the summer of 1954 are entered in table 18. They run from 6638 to 6706. The culture numbers for the summer of 1955 run from

6861 to 6900. Those for the summer of 1956, given in this section, are few, being 7260, 7261, and 7285. Those for the summer of 1957 given in this section likewise are few. They run from 7330 to 7334.

Another point should be made clear at this time. In considering Spm number in a plant, it must be stated that it is not possible to distinguish the presence of two or more Spm elements when these are located very close to one another in a chromosome. Increase in dose of Spm does not effect change in mutation pattern given by the states of a_1^{m-1} that have been isolated. The same pattern of this appears when either one or more Spm elements are present. One state of a_2^{m-1} allows the different doses of Spm to be distinguished from each other but a state of this type was not isolated early in the study of a_1^{m-1} . Therefore, in this section, when a plant is stated to have one Spm, this does not exclude the possibility that two or more Spm elements were present in one of its chromosomes but located so close to each other that detection of this could not be made. The inheritance pattern among the kernels on a testcross ear would be that of one Spm at one particular location in the chromosome complement.

At times, mention will be made of sectors appearing on some ears in which no Spm activity is given by any of the kernels within the sector. Sometimes the boundaries of the sectors were clearly defined and it was possible to ~~knowingly~~ isolate all kernels within it. With others, the kernel types on the ~~pear~~ would not allow the exact boundaries to be determined. However, experience with the forms of sectors appearing on ears allows one to project the boundaries of those sectors that are not accurately defined by kernel phenotype. When such appeared, the boundaries were drawn ^{the} on ^{such} an ear ~~xxx~~ by ^{projections}, and the kernels within the sector removed from the ear and ^{sorted} ~~counted~~ separately. Undoubtedly, some errors were made in projecting the ^a boundaries, but the degree of this could not have been great. Inclusion/ⁱⁿ or exclusion/ of a few kernels ^{seriously} ~~that was present~~ (from the sector) would not/alter the ratio of kernel types on the rest of the ear.

As indicated in table 30, the two Spm elements in plant A-8, and two of the four Spm elements in plant A-2, occupied positions in the Y bearing chromosome⁶/in each. Evidence of this, derived from tests conducted with the progeny of plant A-8, will be summarized.

Progeny Test 2, figure 2

The testcross ear of plant 6629A-8 had the kernel types entered in table 5. The Spm constitution of 19 plants derived from the variegated, Y class of kernels on this ear was determined in the manner described in the previous section. Eleven of these plants were Y Spm/y + in constitution. The other 8 plants had at least 2 Spm elements, each carried in the Y bearing chromosome 6. The constitution of these plants will be symbolized as Y Spm Spm/y + +. Eleven plants derived from the Y class of colorless, sh₂ kernels also were examined. Spm was absent from one of them but present in the other 10 plants. In 5 of the ~~other~~ 10 plants, one Spm was present and it was carried in the Y bearing chromosome 6 in each (Y Spm/y +). In the other 5 plants, more than one Spm was present and in all plants, each of them was located in the Y bearing chromosome 6 (Y Spm Spm/y + +). In addition, thirteen plants derived from the y class of colorless, sh₂ kernels were examined. In 9 of them, no Spm was present. One Spm was present in 3 of the 4 plants having it (line 7, table 31) and two Spm elements were present in the fourth plant (line 8,